

al., U.S. Serial No. 07/882,771, filed May 14, 1992, and now U.S. Patent No. 5,364,791, entitled "Progesterone Receptor Having C Terminal Hormone Binding Domain Truncations." All of these applications and patents (including drawings) are hereby incorporated in their entirety by reference. --

Please incorporate the enclosed paper copy of the Sequence Listing into the specification of the instant application.

### **In The Claims**

Please cancel claims 1-99 without prejudice. Applicant reserves the right to pursue the cancelled subject matter in this or any other appropriate patent application. The cancellation of these claims makes no admission regarding the patentability of this subject matter and should not be so construed.

Please add the following new claims:

100. (New) A method of regulating expression of a desired gene in an animal, plant or cell, said method comprising:

administering to said animal, plant, or cell a pharmacological dose of a ligand which binds to a mutated steroid receptor superfamily ligand binding domain,

wherein said animal, plant, or cell, contains:

(a) a first nucleic acid cassette which comprises a promoter transcriptionally linked to a mutated receptor protein coding sequence,

wherein said mutated receptor protein coding sequence comprises a nucleic acid sequence encoding a mutated receptor protein which regulates the transcription of a molecular switch promoter, and wherein said mutated receptor protein comprises:

a DNA binding domain which binds said molecular switch promoter;

a mutated steroid hormone receptor superfamily ligand binding domain distinct from a naturally occurring ligand binding domain;

a transactivation domain which causes transcription from said molecular switch promoter when said mutated receptor protein is bound to said molecular switch promoter and to an antagonist for a nonmutated receptor protein; and

(b) transcriptionally linked to said molecular switch promoter, a nucleic acid encoding a desired protein in a second nucleic acid cassette; wherein administration of said ligand regulates expression of said desired gene in said animal, plant, or cell.

101. (New) The method of claim 100, wherein the mutated steroid hormone superfamily receptor ligand binding domain is selected from the group consisting of estrogen, androgen, Vitamin D, COUP-TF, cis-retinoic acid, Nurr-1, thyroid hormone, mineralocorticoid, glucocorticoid-alpha, glucocorticoid-beta, and orphan receptor ligand binding domains.

102. (New) The method of claim 100, wherein the mutated receptor protein is comprised of a progesterone receptor with the native DNA binding domain replaced with a GAL-4 DNA binding domain.

103. (New) The method of claim 100, wherein the nucleic acid encoding said desired protein is transcribed to produce an mRNA molecule that is translated to produce a protein after the animal, plant or cell is given a dose of a ligand which binds to the mutated steroid hormone receptor superfamily ligand binding domain.

104. (New) The method of claim 100, wherein the first nucleic acid cassette and the second nucleic acid cassette in said animal, plant, or cell are on separate plasmids.

105. (New) The method of claim 100, wherein the mutated steroid receptor comprises a non-native or modified DNA binding domain.

106. (New) The method of claim 100, wherein said ligand is administered to an animal.
107. (New) The method of claim 106, wherein said animal is a mammal.
108. (New) The method of claim 107, wherein said mammal is a human.
109. (New) The method of claim 100, wherein said ligand is administered to a cell.
110. (New) The method of claim 100, wherein said ligand is administered to a plant.
111. (New) The method of claim 100, wherein the molecular switch is linked to a nucleic acid cassette thereby forming a cassette/molecular switch complex and said complex is positionally and sequentially oriented in a vector such that the nucleic acid in the cassette is transcribed and translated in said target animal, plant, or cell.
112. (New) The method of claim 100, wherein the mutated steroid hormone receptor ligand binding domain includes an ecdysone ligand binding domain.
113. (New) The method of claim 100, wherein the mutated steroid hormone receptor ligand binding domain binds a compound selected from the group consisting of 5-alpha-pregnane-3,2-dione; 11 beta-(4-dimethylaminophenyl)-17 beta-hydroxy-17 alpha-propinyl-4,9-estradiene-3-one; 11 beta-(4-dimethylaminophenyl)-17 alpha-hydroxy-17 beta-(3-hydroxypropyl)-13 alpha-methyl-4,9-gonadiene-3-one; 11 beta-(4-acetylphenyl)-17 beta-hydroxy-17 alpha-(1-propinyl)-4,9-estradiene-3-one; 11 beta-(4-dimethylaminophenyl)-17 beta-hydroxy-17-alpha-(3-hydroxy-1 (Z)-propenyl-estra-4,9-diene-3-one; (7 beta,11 beta,17 beta)-11-

(4-dimethylaminophenyl)-7-methyl-4',5'-dihydrospiro[ester-4,9-diene-17,2'(3'H)-furan]-3-one;  
(11 beta,14 beta,17 alpha)-4',5'-dihydro-11-(4-dimethylaminophenyl)-[spiroestra-4,9-diene-  
17,2'(3'H)-furan]-3-one.

114. (New) The method of claim 100, wherein the mutated steroid hormone superfamily receptor ligand binding domain is mutated to bind a compound selected from the group consisting of non-natural ligands, non-native hormones and anti-hormones.

115. (New) The method of claim 100, wherein said DNA binding domain is replaced with a DNA binding domain selected from the group consisting of GAL-4 DNA binding domain, virus DNA binding domain, insect DNA binding domain and a non-mammalian DNA binding domain.

116. (New) The method of claim 100, wherein said transactivation domain is selected from the group consisting of VP-16, TAF-1, TAF-2, TAU-2.

117. (New) The method of claim 116, wherein said transactivation domain comprises a TAF-1 transactivation domain.

118. (New) The method of claim 100, wherein said transactivation domain is a VP-16 transcription region and wherein said DNA binding domain is a GAL-4 DNA binding domain.

119. (New) The method of claim 100, wherein said transactivation domain is a TAF-1 transcription region and wherein said DNA binding domain is a GAL-4 binding domain.

120. (New) The method of claim 100, wherein said molecular switch is tissue specific.

121. (New) The method of claim 120, wherein the tissue specificity of said molecular switch is controlled by selection of a tissue-specific transactivation domain.
122. (New) The method of claim 120, wherein the molecular switch further comprises a tissue-specific cis-element.
123. (New) The method of claim 100, wherein said mutated steroid receptor results from a deletion in its carboxy terminal amino acids.
124. (New) The method of claim 109, wherein said cell is selected from the group consisting of yeast, mammalian and insect cells.
125. (New) The method of claim 124, wherein said ligand is administered to a mammalian cell.
126. (New) The method of claim 125, wherein said mammalian cell is selected from the group consisting of HeLa, CV-1, COSM6, HepG2, CHO and Ros 17.2.
127. (New) The method of claim 100, wherein said ligand is an endogenous ligand for said mutated steroid hormone receptor.
128. (New) The method of claim 126, wherein said ligand is administered at a dose of about  $10^{-7}$  M to about  $10^{-6}$  M.
129. (New) The method of claim 128, wherein said ligand is 11 beta-(4-dimethylaminophenyl)-17 beta-hydroxy-17 alpha-propinyl-4,9-estradiene-3-one.